

REMARKS

Applicant requests reconsideration of the present application in view of these comments.

I. Status of Claims

Claim 8 has been amended. Support for the amendments can be found at least in the specification at page 9, lines 7-12 and original claims 9 and 16. Meanwhile, claims 9, 16, and 23-30 have been cancelled. Upon entry of the proposed amendments, therefore, claims 8, 10-15, and 17-22 will be pending.

II. Claim Objections

The examiner objects to claim 35 for reciting a “use”. Applicant has cancelled the claim, thus the objection is now moot.

III. Rejections Under 35 U.S.C. § 102(e)

The examiner rejects claims 8-12, 15-19, 21-24, 26-27 and 30 for alleged anticipation by Sabbadini *et al.*, U.S. patent No. 7,183,105 (“Sabbadini”). Applicant respectfully traverses the rejection.

A. Sabbadini Fails to Teach a Method Comprising Each of the Presently Recited Elements

As an initial matter, applicant notes that nowhere does Sabbadini teach a targeted drug delivery method as presently claimed. To characterize Sabbadini as anticipatory, the examiner endeavors to recreate a method as presently claimed by cherry-picking elements from several “laundry list” disclosures in the Sabbadini text and then combining those elements, an approach that Section 102 does not countenance. In other words, the examiner errs factually in contending that Sabbadini makes accessible to the interested public a method that includes each of the presently recited elements, as Section 102 requires of an anticipatory reference. *See, e.g., In re Hall*, 781 F.2d 897, 899, 228 USPQ 453, 455 (Fed.Cir. 1986); *In re Wyer*, 655 F.2d 221, 226-27, 210 USPQ 790, 794-95 (CCPA 1981).

Under the “minicell” rubric, for instance, the Sabbadini text presents a broad genus of cell types, including eubacterial minicells, eukaryotic minicells, and archeabacterial minicells. *See* column 38, line 30 to column 41, line 15. Additionally, Sabbadini’s “eubacterial minicell” category

subsumes a variety of cell types, such as poroplasts, spheroplasts and protoplasts. Column 111, line 54-62.

Another laundry list is evident in Sabbadini's characterization of the minicell as useful for delivering "therapeutic agents", which Sabbadini defines as "any type of compound or moiety". Column 7, lines 7-18. In "non-limiting" fashion, the text lists "small molecules, polypeptides, antibodies, antibody derivatives, nucleic acids, drugs, prodrugs and immunogens" in this context. *Id.*

As for targeting, Sabbadini notes that a minicell may display a "binding moiety". Column 7, lines 9-12. Sabbadini broadly defines such moieties as "any chemical composition, i.e., a small molecule, a nucleic acid, a radioisotope, a lipid or a polypeptide." Column 136, lines 56-63. While Sabbadini identifies an antibody and polyethylene glycol as "non-limiting examples" of binding moieties that can be covalently attached to minicells (Column 136, lines 56-67), nothing in the cited material suggests bringing bispecific ligands into contact with any of Sabbadini's "minicells." For this reason alone, the anticipation rejection must fail.

To create some semblance of the claimed methodology, in any event, the knowledgeable reader would be obliged to pick and choose from among Sabbadini's broad genera, discussed above, even to attempt a permutation that corresponded to the combination of elements recited in applicant's claims, *e.g.*, "targeted drug delivery method," "bispecific ligands having specificity for a mammalian cell surface receptor capable of activating receptor-mediated endocytosis," "intact, bacterially derived minicell," and "small molecule drug." Where, as here, "it is necessary to select portions of teachings within a reference and [to] combine them, ... anticipation can only be found if the [prior disclosed] classes of [items] are sufficiently limited or well delineated." MPEP § 2131.02, *citing Ex parte A*, 17 USPQ2d 1716 (Bd. Pat. App. & Inter. 1990).

The teachings of Sabbadini are anything but limited or well-delineated, as applicant has demonstrated. One of ordinary skill certainly could not "at once envisage" in Sabbadini's text the prescribed combination of applicant's invention. Yet this is what the PTO's own rules require for anticipation in this context. *Id.*, *citing In re Petering*, 301 F.2d 676, 133 USPQ 275 (CCPA 1962).

Moreover, an artisan picking and choosing from among Sabbadini's broad genera would have been dissuaded from even pursuing a method involving the recited combination of elements. As applicant's disclosure underscores, for example, at page 3, lines 8-16, certain events must occur before a bacterially-derived recombinant minicell can deliver, to a targeted non-phagocytic

mammalian cell, a small molecule drug in accordance with the claimed invention. As an initial matter, (1) the recombinant minicell must recognize the target non-phagocytic mammalian cell specifically and (2) the recombinant minicell must be internalized into the target non-phagocytic mammalian cell. Conventional wisdom, however, held that large particles like intact bacterially derived minicells could not passively enter non-phagocytic mammalian cells via receptor-mediated endocytosis. In view of the lack of direction provided by Sabbadini and the absence of any working example, a practitioner would have been dissuaded from even attempting to deliver a small molecule drug to non-phagocytic mammalian cells via bispecific ligands having specificity for a mammalian cell surface receptor capable of activating receptor-mediated endocytosis. Any attempt to depart from conventional wisdom and employ such a method, moreover, would have required extensive research.

B. Conventional wisdom held that large particles like intact bacterially derived could not passively enter non-phagocytic mammalian cells via receptor-mediated endocytosis

The skilled artisan had no reason to have expected success in achieving events 1-2, *supra*, with minicell vectors. Although ligands had reportedly been used to bring a gene delivery vector into contact with a desired non-phagocytic mammalian cell, that concept cannot be generalized to all types of drug delivery vectors, particularly to minicell vectors.

For example, adenoviral vectors have been redirected to target mammalian cell-surface receptors, such as endoglin on endothelial cells, and internalized via clathrin-coated pits in the mammalian cell plasma membrane. Wickham *et al.*, 1996; Nettelbeck *et al.*, 2001; Boucher *et al.*, 2003. The clathrin-coated pits resemble a cup that envelopes the vector, but the size of the cup is understood to be a limiting factor. Clathrin-coated pits have a limited size of 85-110 nm, due to the size of the clathrin coat. Swanson & Watts, 1995. Minicells are at least 400 nm in diameter, by contrast; hence, the skilled artisan would not have expected this targeting approach to work for minicells.

Knowledge concerning other large vectors further supported the expectation that minicells would not be internalized through clathrin-coated pits. For instance, large lipoplexes (non-viral vectors up to 500 nm) preferentially enter cells by receptor- and clathrin-independent endocytosis, while smaller lipoplexes (less than 200 nm) can be internalized via a non-specific, clathrin-dependent process. Simoes *et al.*, 1999. Likewise, large viruses, such as vaccinia

virus, on the order of 350 nm x 250 nm in size do not infect mammalian cells via a clathrin-coated pathway. Essani and Dales, 1979.

In a similar vein, it was known that non-phagocytic mammalian cells cannot engulf large pathogens, like bacterial cells. Only professional phagocytes like macrophages engulf such pathogens, and the engulfment process is clathrin- and receptor-independent, being accomplished by phagocytosis. The interaction of large pathogens with the cell surface induces a complex signaling cascade, leading to actin rearrangements at the plasma membrane to form a large phagocytic cup, which engulfs the bacterium. Dramsi and Cossart, 1998. Furthermore, artisans at the time had only a rudimentary understanding of the signaling cascades responsible, on bacterial entry, for actin rearrangements at the plasma membrane. Galan, 1996; Menard *et al.*, 1996; Finlay and Cossart, 1997; Dramsi and Cossart, 1998.

Specific investigations into the effect of particle size on receptor-mediated endocytosis showed the process to be strongly size-dependent. For example, Aoyama *et al.*, 2003, studied the effect of particle size on glycoviral gene delivery and concluded that the optimal particle size for receptor-mediated endocytosis is ~25 nm. *See also* Nakai *et al.*, 2003; Osaki *et al.*, 2004. Gao *et al.*, 2005, confirmed that conclusion.

Accordingly, conventional wisdom held that large particles like intact bacterially derived minicells (*i.e.*, approximately 400 nm) could not passively enter non-phagocytic mammalian cells via receptor-mediated endocytosis.

C. Sabbadini Provides Insufficient Guidance for the Skilled Person to Effect Targeted Drug Delivery, as Claimed

In line with conventional thinking, Sabbadini, in the cited section on “Cancer Therapy”, suggests that macrophages are required to accomplish receptor-mediated endocytosis and that for direct access into target cells, expression of a toxR-invasin fusion protein might be used to an actively induce entry. See column 171, line 53 to column 172, line 3. Similarly, Sabbadini in cited example 19 advocates for i) using protoplasts, (ii) targeting via expression of antibody fusion constructs and (iii) effecting uptake via forced entry using an active invasive process. *Id.* at Example 19.

From this perspective, it is not surprising that nothing in the cited material propounds a rationale for combining the claimed elements in the recited fashion. Moreover, it must be emphasized that Sabbadini lacks any supporting data or working examples of targeted drug delivery

of any kind. In view of such a hollow disclosure and the conventional wisdom at the time of the invention, the skilled person would have been unable to effect targeted drug delivery as presently claimed.

Accordingly, for these reasons the rejection under Section 102(e) should be withdrawn.

IV. Rejections Under 35 U.S.C. § 103

The examiner rejects claims 8, 11, 13-14 for allegedly being unpatentable over Sabbadini in view of Nettelbeck and Coldwell. The examiner also rejects claims 8, 20, 23 and 25 for allegedly being unpatentable over Sabbadini in view of WO 96/026715 (Hope *et al.*). Applicant respectfully traverses the rejections.

Sabbadini's musings are discussed above. Nettelbeck is cited for allegedly teaching a recombinant antibody as a molecular bridge and the construction and use of a bispecific single chain diabody, and Coldwell is cited for allegedly teaching production of monoclonal antibodies to antigenic determinants of the O-polysaccharide of *Salmonella typhimurium* lipopolysaccharide. Meanwhile, Hope is cited for allegedly teaching a method of loading a chemotherapeutic agent into a liposome. None of these references, however, alone or in combination, cure Sabbadini's deficiencies. Thus, for the reasons expressed above, a practitioner would have been dissuaded from even attempting to use intact, bacterially derived minicells to deliver a small molecule drug to non-phagocytic mammalian cells via bispecific ligands having specificity for a mammalian cell surface receptor capable of activating receptor-mediated endocytosis. Thus, nothing in the record provides the requisite motivation for combining the recited elements in the posited fashion.

Similarly, for the reasons noted above, the skilled person reviewing Sabbadini and the secondary references would not have reasonably expected intact, bacterially derived minicells to passively enter non-phagocytic mammalian cells via receptor-mediated endocytosis. Nor was it expected that small molecule drugs could be packaged into such minicells, another obstacle unaddressed by Sabbadini. In this regard, it was surprising that intact minicell membranes are permeable to a range of structurally dissimilar hydrophilic, hydrophobic and amphipatic drugs. By contrast, live bacterial cells exhibit selective membrane permeability to solutes, suggesting that that minicells have lost this selectivity. It also was surprising that minicells are unable to expel drugs from their cytoplasm because live bacterial cells extrude noxious chemicals that enter into the bacterial cytoplasm. Even against a reverse osmotic gradient, in which drug-loaded minicells are suspended in phosphate-buffered saline containing no drug, minicells retain drug. This was

additionally surprising because drugs appear simply to diffuse into minicells through intact minicell membranes, yet the diffusion channels are not available for drugs to diffuse out of minicells.

Likewise, it was unexpected that therapeutically significant concentrations of drug could be packaged within minicells. As bacterial cytoplasm (and, hence, minicell cytoplasm) contains significant concentrations of biocompatible solutes, it was believed that there was insufficient intracellular space to accommodate high concentrations of non-biocompatible drug solutes, without loss of minicell integrity.

The ability of minicells to deliver drugs also was surprising. It was unexpected, for example, that drug-packaged minicells do not leak drug into the extracellular space. By comparison, this is a persistent problem with liposomal drug delivery vectors, and minicells, like liposomes, are non-living vesicles. Nevertheless, although intact minicell membranes lack selectivity to drug permeation, the membrane integrity is sufficient to prevent leakage of intracellular solutes. Also surprising, and unlike liposomal drug delivery vectors, attachment of ligands to the surface of drug-packaged minicells does not cause destabilization of minicell integrity or membrane perturbations that result in drug leakage.

Yet another surprise was that drug-packaged minicells carrying a bispecific ligand are able to extravasate tumor neovasculature *in vivo*. While there is considerable debate regarding the leakiness of tumor microenvironment neovasculature, the current view is that pores in the neovasculature are 150-400 nm in diameter (Gabizon *et al.*, 2003). Minicells carrying a surface ligand, however, are 400nm to 600nm in diameter, yet still are able to extravasate tumor neovasculature *in-vivo*.

The ability of drugs packaged in minicells to avoid degradation also was surprising. Engulfed minicells are subjected to lysosomal and late-endosomal environments known to be harsh, and which break down minicells. Despite the harshness of these environments, applicant has shown that a range of drugs are released from minicells in a biologically active form and remain significantly unaltered. Perhaps even more surprising was the discovery that a significant concentration of drug is able to escape, in its active form, into the mammalian cell cytoplasm. Pursuant to the present invention, that is, drug concentrations within mammalian cells are sufficient to work a therapeutic effect both *in vitro* and *in vivo*.

Thus, a practitioner at the time of the invention would have lacked a reasonable expectation that intact, bacterially derived minicells could deliver a small molecule drug to non-phagocytic

mammalian cells via bispecific ligands having specificity for a mammalian cell surface receptor capable of activating receptor-mediated endocytosis.

For these reasons, a *prima facie* case of obviousness has not been established. On this basis alone, therefore, the rejections should be withdrawn.

Yet, applicant provides still more evidence of the unobviousness of the claimed methods. In this regard, applicant notes that even today, more than five years after the priority filing, the field remains skeptical of the functionality of the claimed methods to achieve targeted drug delivery. The prevailing view today is still that large particles, like intact bacterially derived minicells, are too big to utilize receptor-mediated endocytosis and must use an active invasive process to induce entry in cells. *See, e.g.* Cossart and Velga (2008); Doherty and McMahon (2009). From this perspective, it is not surprising that applicant's work recently captured the cover of the prestigious journal *Nature Biotech* and was highlighted in *Nature Reviews Cancer*. *See* MacDiarmid *et al.* (2009); McCarthy (2009), which are enclosed.

It is clear, therefore, that the cited combination fails to render the claimed invention obvious. Accordingly, the rejection should be withdrawn.

V. Rejections Under Double Patenting

The examiner provisionally rejects the pending claims under obviousness-type double patenting over U.S. application No. 11/765,635. While acknowledging this ground of rejection, applicant requests that the examiner hold it in abeyance until such time as he indicates allowable subject matter. Should a concern remain, then applicant will address the merits of the rejection.

Applicant requests an early indication that this application is in allowable condition. Examiner Singh is invited to contact the undersigned directly should he feel that any issue requires further consideration.

The Commissioner is hereby authorized to charge any additional fee, which may be required under 37 C.F.R. §§ 1.16-1.17, and to credit any overpayment to Deposit Account No. 19-0741. Should no proper payment accompany this response, then the Commissioner is authorized to charge the unpaid amount to the same deposit account. If any extension is needed for timely acceptance of

submitted papers, then applicant petitions for such extension under 37 C.F.R. §1.136 and authorizes payment of the relevant fee from the deposit account.

Respectfully submitted,

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By R. Brian McCaslin

FOLEY & LARDNER LLP
Customer Number: 22428
Telephone: (617) 342-4039
Facsimile: (617) 342-4001

R. Brian McCaslin
Attorney for Applicant
Registration No. 48,571

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